REMARKS

The Applicants have replaced claims 1-26, drawn to pharmaceutical compositions or diagnostic kits with claims 27-34, drawn to a method for protecting dopaminergic neurons. In the remarks below, the Applicants address all of the Examiner's objections and rejections and believe they are overcome by a combination of the amendments and remarks presented herein. Reconsideration of the application is respectfully requested.

The Specification has been amended as requested by the Examiner to identify the remaining unidentified sequences with sequence identifiers and to correct informalities in the presentation of the descriptions of the figures.

New claims 27-34 have also been written so as to correct the objected-to informalities identified by the Examiner in now-cancelled claims 1-26.

Rejections for Indefiniteness

New claims 27-34 have been written so as to address the indefiniteness matters pointed out by the Examiner in rejection nos. 10-13.

As to rejection no. 14, regarding the asserted lack of enablement concerning the claimed treatment of "disorders characterized by a degeneration of dopaminergic neurons," it should be noted that in stroke, Parkinson's disease, Alzheimer's disease, dementias, and infections of the central nervous system, a more or less severe degeneration of dopaminergic neurons is observed in addition to other damages of the brain, which often have an unclear origin. Together, degeneration of dopaminergic neurons and damages of the brain contribute to the overall picture of the disease. Although it is well understood

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by those skilled in the art that a variety of types of neuronal cells are affected in the above disorders and diseases, *there is always a loss of dopaminergic cells*, and the reduction of this loss lessens the lasting damages caused by these disorders and diseases.

In order to demonstrate that this is common knowledge to those skilled in the art, the following documents are enclosed and referenced herein as follows:

Referenced documents

1. Zhang et al., "Parkinson's Disease is Associated with Oxidative Damage to Cytoplasmic DNA and RNA in Substantia Nigra Neurons", American Journal of Pathology, Vol. 154, No. 5, pp. 1423-1429, May 1999.

2. Yoo et al., "Oxidative Stress Regulated Genes in Nigral Dopaminergic Neuronal Cells: Correlation with the Known Pathology in Parkinson's Disease", Brain Research Mol. Brian Res., Vol. 110, No. 1, pp. 76-84, January 2003.

3. Kazee et al., "Substantia nigra lesions in Alzheimer Disease and Normal Aging", Vol. 9, No. 2, pp. 61-67, Summer 1995.

4. Kastner et al., "Immunocytochemical Quantification of Tyrosine Hydroxylase at a Cellular Level in the Mesencephalon of Control Subjects and Patients with Parkinson's and Alzheimer's Disease", J. Neurochem, Vol 61, No. 3, pp. 1024-1034, September 1993.

5. Oliver et al., "Specific Infection and Destruction of Dopaminergic Neurons in the Substantia Nigra by Theiler's Virus", Journal of Virology, Vol. 71, No. 8, pp. 6179-6182, August 1997.

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6. Nath et al., "Neurotoxicity and Dysfunction of Dopaminergic Systems Associated with AIDS Dementia", J. Psychopharmacol., Vol. 14, No. 3, pp. 222-227, 2000.

7. Yun et al., "Extensive Degeneration of Catecholaminergic Neurons to Scrapie Agent 87V in the Brains of IM Mice", Mol. Chem. Neuropathol., Vol. 34, No. 2-3, pp. 121-132, Jun-Aug 1998.

8. Verity et al., "Mesolimbocortical Dementia: Clinico-Pathological Studies of Two Cases", Vol 53, No. 6, pp. 492-495, June 1990.

9. Perry et al., "Alteration in Nicotine Binding Sites in Parkinson's Disease, Lewy Body Dementia and Alzheimer's Disease: Possible Index of Early Neuropathology", Neuroscience, Vol. 64, No. 2, pp. 385-395, January 1995.

10. Thomas et al., "In Vitro and In Vivo Activity of a Novel Series of Radical Trapping Agents in Model Systems of CNS Oxidative Damage", Ann. NY Acad. Sci., Vol. 17, No. 738, pp. 243-249, November 1994.

11. Jain et al., "Matrix Metalloproteinases and Free Radicals in Cerebral Ischemia", Free Radic. Biol. Med., Vol. 39, No. 1, pp. 71-80, July 2005.

Parkinson's disease (PD)

(1) Zhang et al. 1999 (cf. for example page 1423, column 2):

"The causes of dopaminergic neurodegeneration in PD remain unclear, but several lines of evidence suggest involvement of oxidative stress in PD pathogenesis. First, PD is associated with both increased levels of nigral iron, a catalytic agent for production of hydroxyl radical, and increased Mn superoxide

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dismutase activity. Second, midbrain levels of reduced glutathione are diminished in PD patients. Third, there is evidence of increased oxidative damage in midbrain from PD patients, including lipid peroxidation, protein oxidation, and oxidation of DNA. Finally, several laboratories have observed increased catechol oxidation in the midbrain of PD patients." (Emphasis added; footnotes omitted)

(2) Yoo et al. 2003 (Abstract):

"Oxidative stress (OS) is a primary pathogenic mechanism of nigral dopaminergic (DA) cell death in Parkinson's disease (PD). Oxidative damage, Lewy body formation and decreased mitochondrial complex I activity are the consistent pathological findings in PD."

Alzheimer disease (AD)

In this context, it should be noted that the substantia nigra is a region of brain containing dopaminergic neurons, particularly pigmented dopaminergic neurons.

(3) Kazee et al. 1995 (Abstract):

"Clinical and pathological overlap between Alzheimer disease (AD) and Parkinson's disease (PD) has been well described; however, the mechanisms of overlap between these two disorders remain unknown. We retrospectively examined clinical and neuropathological features from 66 individuals participating in the Rochester Alzheimer Disease Center to determine the

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association of AD with substantia nigra (SN) pathology. SN pathology, identified by a loss of pigmented neurons and the presence of gliosis, pigment-laden macrophages, and Lewy bodies, was blindly scored in 48 AD cases and 18 normal elderly controls. We found moderate or severe pathology in the SN in 2 control brains (11%) and 29 AD brains (60%). The numbers of neocortical and hippocampal neurofibrillary tangles (NFTs) and senile plaques (SPs) were not associated with nigral pathology. There was also no significant association of SN pathology with NFTs or SPs in the striatum, the site to which these neurons project. There was no significant association of increasing SN pathology with aging among AD patients, nor with increasing severity and duration of AD. The signs and symptoms of an extrapyramidal movement disorder were, however, associated with increasing SN pathology. We confirm that pathological lesions in the SN are a common feature of AD and an uncommon feature in normal aging. AD is a significant risk factor for SN lesions and PD, but the pathologic severity of AD, as measured by NFTs and SPs, was not associated with SN lesions." (Emphasis added)

(4) Kastner et al. 1993 (Abstract):

"In patients with Alzheimer's disease, the amount of TH was selectively reduced in the remaining dopaminergic neurons of the ventral tegmental area, a region characterized by a loss in dopaminergic neurons. The decrease in cellular TH content might therefore be related to the presence of the neurodegenerative process in the area considered." (Emphasis added)

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Infections of the CNS

(5) Oliver et al. 1997 report the selective destruction of dopaminergic neurons in the substantia nigra after an infection with Theiler's Virus (Abstract):

"Theiler's murine encephalomyelitis virus was stereotaxically inoculated unilaterally into the substantia nigra of the mouse brain. Virus specifically infected tyrosine hydroxylase-positive neurons and spread rostrocaudally throughout this subpopulation of neurons, resulting in impaired function and degeneration of the substantia nigra. The spread of the virus to other areas of the brain was minimal and rare." (Emphasis added)

(6) Nath et al. 2000 (Abstract):

"Infection with the human immunodeficiency virus (HIV) selectively targets the basal ganglia resulting in loss of dopaminergic neurons. Although frequently asymptomatic, some patients may develop signs of dopamine deficiency de novo. Accordingly, they are highly susceptible to drugs that act on dopaminergic systems. Both neuroleptics and psychostimulants may exacerbate these symptoms. Experimental evidence suggests that viral proteins such as gp120 and Tat can cause toxicity to dopaminergic neurons, and this toxicity is synergistic with compounds such as methamphetamine and cocaine that also act on the dopaminergic system." (Emphasis added)

(7) Yun et al. 1998 (Abstract):

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"Scrapie is a degenerative disease of the central nervous system of sheep and goats. The causative agent has been passaged to a number of laboratory species, including mice and hamster. Amyloid plaque formation and vacuolation, the signs of senile dementia, are found in the brains of mice infected with 87V scrapie agent. Dopamine (DA) and norepinephrine (NE) concentrations in the brains of scrapie-infected mice were measured with high-performance liquid chromatography-electrochemical detector (HPLC-ECD). A significant decrease in NE level was exhibited in all regions tested, whereas the level of DA decreased significantly only in cerebral cortex. Immunohistochemistry was used to examine immunoreactive catecholamine neurons in substantia nigra and locus ceruleus using antisera against tyrosine hydroxylase (TH). The population of TH-immunoreactive neurons in the substantia nigra and locus ceruleus were significantly decreased in scrapie-infected mice compared to controls. These data suggest that both the noradrenergic and dopaminergic system are sensitive to the action of scrapie agent 87V and that changes in the catecholamine levels in the brains of scrapie-infected mice may contribute to some of the clinical symptoms of the diseases, such as ataxia and apraxia."

Dementia

(8) Verity et al. 1990 (Abstract):

"The clinicopathological findings are presented of two cases of mesolimbocortical dementia. Both cases were characterised by late onset slowly progressive personality changes and

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progressive intellectual deterioration without clinical Parkinsonism. Neuropathological findings revealed non-specific neuronal degeneration, Holzer and GFAP positive gliosis primarily affecting the limbic system, caudate, thalamus and substantia nigra. The pathological findings coincide with the distribution of the non-striatal dopaminergic pathways and suggest an intrinsic involvement of these pathways to account for the clinical and pathological manifestations." (Emphasis added)

(9) Perry et al. 1995 (Abstract):

"High-affinity nicotine binding, considered to primarily reflect the presence of CNS alpha 4 beta 2 nicotinic receptor subunits, was examined autoradiographically in brain regions most severely affected by Alzheimer and Parkinson types of pathology. In the midbrain, the high density of binding associated with the pars compacta of the substantia nigra was extensively reduced (65-75%, particularly in the lateral portion) in both Lewy body dementia and Parkinson's disease. Since loss of dopaminergic neurons in Lewy body dementia was only moderate (40%), loss or down-regulation of the nicotinic receptor may precede degeneration of dopaminergic neurons in this region….Abnormalities of the nicotinic receptor in the diseases examined appear to be closely associated with primary histopathological changes: dopaminergic cell loss in Parkinson's disease and Lewy body dementia, amyloid plaques and tangles in subicular and entorhinal areas in Alzheimer's disease. Loss or down-regulation of the receptor may precede neurodegeneration." (Emphasis added)

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Stroke/cerebral ischemia

A stroke results in damages of the CNS including damages of dopaminergic neurons. The following citations show that said damages have at least partially an oxidative origin and thus can be treated with GDF-15 according to the results given in the examples of the present application.

(10) Thomas et al. 1994 (Abstract):

"Many laboratory and clinical studies suggest that oxygen radical formation and resultant cell damage contribute to CNS injury following stroke and neurotrauma. Accordingly, antioxidants represent a viable therapeutic approach for management of CNS oxidative damage. Recently, several investigators have reported that the spin trap PBN protects against stroked-induced damage and reduces aging-associated neurological deficits. We have prepared and tested a cyclic analog of PBN, MDL 101,002, in a number of in vitro and in vivo assays designed to assess its neuroprotective properties. MDL 101,002 was found to be an effective .OH trap, to inhibit lipid peroxidation, and to decrease infarct size in a gerbil model of stroke. These results further indicate that oxidative damage arising from stroke contributes to infarct formation, and that spin traps are effective in ameliorating ischemia and reperfusion-induced CNS injury." (Emphasis added)

(11) Jian et al. 2005 (Abstract):

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"Cerebral ischemia induces a complex series of molecular pathways involving signaling mechanisms, gene transcription, and protein formation. The proteases and free radicals involved are important, both individually and in concert, at each of the steps in the injury cascade. Matrix metalloproteinases (MMPs) and serine proteases are essential in the breakdown of the extracellular matrix around cerebral blood vessels and neurons, and their action leads to opening of the blood-brain barrier, brain edema, hemorrhage, and cell death. Reactive oxygen and nitrogen species affect the signaling pathways that induce the enzymes, the stability of the mRNA, and their activation processes. Mice that either lack MMP genes or overexpress free radical-removing genes exhibit diminished cerebral damage after stroke. Drugs that block MMP activity, or are free radical scavengers, significantly reduce ischemic damage. Understanding the relationship between proteases and free radicals in cerebral ischemia is critical for the design of therapeutic agents aimed at controlling cell death in ischemic tissues." (Emphasis added)

Regarding the objections raised by the Examiner under rejection no. 23 of the Office Action, it should be noted that the experiments outlined in the present application are in accordance with generally recognized model systems for oxidative and toxic influences of dopaminergic neurons which are sufficient to predict the likelihood of a positive effect on human disorders associated with damaged dopaminergic neurons. This fact is emphasized by the affidavit of Prof. Dr. Unsicker enclosed herewith.

The present application provides *inter alia* a new method for the treatment of disorders in mammals that are characterized by

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a degeneration of dopaminergic neurons and discloses the agents needed for carrying out this method. All additionally required experiments are routine experiments that can be carried out by a person skilled in the art. Further, the additional experiments do not require any inventive step and have been carried out routinely with innumerable agents.

Thus, the Applicants assert that the subject matter of new claims 27 to 34 is disclosed sufficiently clearly and concisely for it to be carried out by a person skilled in the art.

Rejections over the Prior Art

Referring to rejection nos. 26 to 33 of the Office Action, it should be noted that none of the cited prior art documents discloses or suggests a method for protecting dopaminergic neurons against neuronal degeneration. In particular, there is no hint given in the documents cited by the Examiner towards a method as defined in new claim 27.

Therefore, the subject matter of new claim 27 is not only novel vis-à-vis the cited prior art documents but also involves an inventive step in view of these documents. The same applies *mutatis mutandis* to the subject matter of new claims 28 to 34, which are either directly or indirectly dependent thereon.

Thus, the Applicants submit that all claims in the application are in condition for allowance and such action is requested.

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The Examiner is encouraged to telephone the undersigned attorney to discuss any matter that would expedite allowance of the present application.

Respectfully submitted,

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